

REMARKS/ARGUMENTS

Status of the Claims

Claims 1 to 12 and 32 to 47 were previously pending and undergoing examination. Claims 1, 6, 9 to 12, 36 to 38, 41, and 47 are currently amended. Claims 48 to 71 are new. Claims 4, 5, 13 to 35, and 39 to 42 are canceled without prejudice. After entry of these amendments, claims 1 to 3, 7 to 12, 36 to 38, 41, and 43 to 71 will be pending.

Claims 1 to 12 and 32 to 47 stand rejected for alleged non-statutory obviousness type double patenting over U.S. Patent No. 6,746,870.

Claims 1 to 12 and 36 to 47 stand rejected under 35 U.S.C. §101 as allegedly drawn to unpatentable subject matter.

Claims 1 to 12 and 36 to 43 stand rejected under U.S.C. §112, first paragraph as allegedly lacking enablement commensurate with the claims.

Claims 9 and 35 stand rejected for allegedly failing to deposit essential biological material.

Claims 1 to 12 and 32 to 47 stand rejected under 35 U.S.C. §112, first paragraph, for allegedly failing to comply with the written description requirement.

Claims 36, 37 and 42 to 47 stand rejected under 35 U.S.C. §112, second paragraph, as allegedly being indefinite.

Claims 1, 2, 4 to 8, 10 to 12, and 32 to 34 stand rejected under 35 U.S.C. §102 (e) as allegedly being anticipated by Crouzet et al. (U.S. Patent No. 6,143,530).

Claims 1 to 12, 32 to 34 and 36 to 47 stand rejected under 35 U.S.C. §103(a) as being unpatentable over Crouzet and further in view of Thorpe and Smith (PNAS 95:5505-5510 (1998)).

Support for the Amendments to the Claims

Claim 1 was amended to set forth a plant or yeast eukaryotic cell, *attB* and *attP* recombination sites, and the recombinase is not capable of mediating recombination between an *attL* and *attR* site. Support for this subject matter is found *inter alia* respectively in original claim 10, original claim 4, and original claim 1.

Claim 9 was amended to set forth that the nucleic acid encodes Φ C31 integrase. Support for this subject matter can be found *inter alia* in original claim 3, Figure 1B, and at column 18, line 47.

Claims 10 and 11 were amended, respectively, to set forth yeast and plant cells, respectively. Support for this subject matter is found *inter alia* in original claim 10.

Claim 36 was amended to set forth *attB* and *attP* recombination site and their relative location. Support for such is found in the specification *inter alia* in Figures 3 and 4. Support for the recital of heterologous nucleic acid is found at line 10 on page 6. Support for the plant and yeast subject matter is as set forth above.

Support for the amendment to claims 37, 49, 51, 60, which recite "transgene" is found *inter alia* on page 6, line 10 and the first full paragraph on page 11.

Support for the amendment to claim 38 is found *inter alia* in the previous version of the claim.

Support for the amendment to claim 47 is as set forth for claim 11.

New claims 48 and 50 set forth heterologous nucleic acid subject matter. Support for this subject matter is found in the paragraph *inter alia* at page 6, line 10.

Support for new claims 52 and 53 is set forth, respectively, as for claims 36 and 1 above. Support for the "in culture" subject matter is found *inter alia* in the specification at page 19, line 9.

Support for the subject matter of new claims 54 and 55 can be found *inter alia* in original claims 10 and 11.

Support for the subject matter of new claim 56 and 57, can be found *inter alia* in original claim 2.

Support for the non-human subject matter of new claim 58 can be found *inter alia* in the specification at page 14, first full paragraph and at page 22, line 4.

Support for the subject matter of new claim 59 can be found *inter alia* in the first full paragraph at page 23.

Support for the subject matter of new claims 61 to 71 can be found *inter alia* in original claims 13 to 22 which were previously canceled without prejudice and in original claim 4 which set forth *attB* and *attP*.

In view of the above, the Applicants believe the amendments to the claims add no new matter and respectfully request their entry.

Status of the Priority Claim

The Applicants filed a Preliminary Amendment with the Application filing papers. The Preliminary Amendment sets forth that the first sentence of the specification is to be amended to set forth the priority claim as correctly recited on the Filing Receipt. Applicants also submit herewith an ADS which sets forth the priority claim recited in the Preliminary Amendment and on the Filing Receipt.

Accordingly, the Applicants believe the intended priority claim is perfected.

Response to the Rejection of Claims 1 to 12 and 32 to 47 for alleged non-statutory obviousness type double patenting over U.S. Patent No. 6,746,870.

The Applicants respectfully request that this grounds for rejection be held in abeyance until such time as the application is otherwise deemed to be in condition for an allowance. At such time, if still required, the Applicants intend to provide a suitable terminal disclaimer.

Response to the rejection of claims 1 to 12 and 36 to 47 under 35 U.S.C. §101 for allegedly claiming unpatentable subject matter

The Applicants present composition base claims which set forth eukaryotic cells that are either "non-human" (claim 58), "plant or yeast" (claim 1 and 36), or "in culture" (claims 52 and 53). Accordingly, the Applicants respectfully request that the above rejection be reconsidered and withdrawn.

Response to the rejection of claims 9 and 35 for an alleged failure to deposit essential biological material.

Without acquiescing on the merits and in order to facilitate examination of the application, the Applicants have amended claim 9 to avoid the recital of the specific plasmid. Claim 35 is canceled without prejudice. Accordingly, the Applicants respectfully request that the above rejections be reconsidered and withdrawn.

Response to the rejection of claims 1 to 12 and 36 to 43 as allegedly lacking enablement commensurate with the claims.

The rejection was largely directed toward subject matter related to the enablement of transgenic animals. Base composition claims 1 and 36 set forth plants. New base composition claims 52 and 53 set forth eukaryotic cells in culture.

Accordingly, the Applicants respectfully request that the above rejections be reconsidered and withdrawn.

Response to the rejection of claims 1 to 12 and 32 to 47 for alleged failure to comply with the written description requirement.

A. Response to the alleged lack of adequate written description with respect to "hybrid recombinase recombination sites."

Without acquiescing on the merits, and in order to facilitate examination of the application, the Applicants have amended the claims to set forth *attR* and *attL* recombination sites.

B. Response to the alleged lack of adequate written description of "the additional factor."

Without acquiescing on the merits, and in order to facilitate examination of the application, the Applicants have amended the claims to avoid reciting "the additional factor." The specification at page 11, lines 20 to 24, explains that eukaryotic cells do not make the additional factor as it is produced by the bacteriophage from which the recombinase is obtained.

Accordingly, the Applicants respectfully request that the above rejections be reconsidered and withdrawn.

Response to the rejection of claims 36, 37 and 42 to 47 for alleged indefiniteness.

Claim 36 was considered to be indefinite for setting forth a *first bacteriophage φC31 recombination site*. Without acquiescing on the merits and in order to expedite prosecution of the instant application, the Applicants have amended the claims to eliminate the phrase at issue. Accordingly, the Applicants respectfully request that the above rejection be reconsidered and withdrawn.

Response to the rejection of claims 1, 2, 4 to 8, 10 to 12, and 32 to 34 for alleged anticipation by Crouzet et al. (U.S. Patent No. 6,143,530, hereinafter, "Crouzet").

Pursuant to MPEP §2131, in order to anticipate a claim, the reference must teach every element of the claim. Applicants respectfully traverse the above rejections on the grounds that Crouzet does not teach every element of the rejected claims. In addition, an alleged anticipatory reference must be enabled with respect to the subject matter at issue. A patent claim "cannot be anticipated by a prior art reference if the allegedly anticipatory disclosures cited as prior art are not enabled." *Elan Pharm., Inc. v. Mayo Found. for Med. Educ. & Research*, 346 F.3d 1051, 1054 (Fed. Cir. 2003).

The Action asserts that Crouzet discloses a eukaryotic cell comprising a prokaryotic recombinase or a construct introduced into the cell. In support of this proposition the Action cites Crouzet at col. 7, lines 45 to 67 and at col. 5, lines 30 to 40. Applicants note that these sections of the Crouzet specification concern methods of producing the Crouzet minicircles in host cells.

The Applicants respectfully disagree with the Action's interpretation of the reference. No where can Crouzet be fairly interpreted as teaching a *prokaryotic* recombinase or construct may be introduced into a *eukaryotic* host cell. Even when mentioning the LoxP system, Crouzet discusses use of a *bacterial* origin of replication (see, column 5, line 61).

Crouzet *only* generally discloses prokaryotic and eukaryotic recombinases may be used in the production of the minicircles of their invention. The Crouzet et al. reference also *only* generally discloses that the methods for producing minicircles can be conducted in bacterial or eukaryotic cells. Crouzet addresses the possible variety of host cells at col. 9, starting at line 47. Such general disclosures would not be fairly construed by one of ordinary skill (for reasons of lack of enablement of such broad interpretation as discussed further immediately below) in the art as *generally* disclosing or suggesting *prokaryotic* recombinases would be suitable for use in the production of the minicircles within *eukaryotic* cells.

In addition, Crouzet further specifies the host cells be *competent* cell hosts with respect to the plasmid and excision (see, column 4 lines 17 to 20). However, Crouzet does not disclose or teach *any* eukaryotic cell as being a competent host with respect to prokaryotic recombinases.

The operability of the Applicants' claimed subject matter in eukaryotes is surprising and one of ordinary skill in the art would not fairly construe Crouzet as teaching or enabling such subject matter. Prokaryotic chromosomes differ from eukaryotic chromosomes in the composition and assembly of different DNA bound proteins, e.g., histones. **Therefore, one of ordinary skill in the art would not generally expect an enzyme recognizing DNA in a prokaryotic cell, or *in vitro*, to operate in a eukaryotic cell.** Evidence as to the inability to obtain function of a prokaryotic enzyme in a eukaryotic cell is therefore an unsurprisingly negative result that is generally not publishable material. However, in instances where at least partial function was obtained, results have been published. These publications provide evidence of an active search for such prokaryotic enzymes that function in the eukaryotic cell and the limited transferability of the prokaryotic activity to the eukaryotic cell.

A most recent example is the expression RecA in plants (Reiss et al., PNAS 97:3358-3363, 2000)(enclosed with IDS) which was published well after the claimed priority date of July 23, 1999. In bacteria, this protein is responsible for homologous recombination and gene targeting. However, despite numerous attempts, only partial success was obtained. Reiss et al. (2000) showed that despite stimulation in intrachromosomal homologous recombination,

specifically sister chromatid exchange and double-strand break repair, the bacterial RecA protein cannot increase the frequency of intermolecular gene targeting in plants.

Another example is the bacteriophage T7 polymerase, where it was expressed in yeast (Benton et al., Molecular and Cellular Biology 10: 353-360, 1990, enclosed with IDS). After adding nuclear import signals to the protein, it was capable of entering the yeast nucleus to direct the transcription of transgenes controlled by the T7 promoter. However, for unknown reasons, those RNA transcripts were not functional. That is, they were not translated, suggesting that the T7 RNA polymerase-directed transcription process in yeast was not faithful.

Similarly, only a few prokaryotic recombination systems have been reported to operate even to a limited extent in a eukaryotic cell. Of the hundreds of site-specific recombination systems known, only a relative few prokaryotic systems are known to work in eukaryotes. Aside from the phiC31 system described here, the others are (see the specification at p.3): the Cre-lox system from bacteriophage P1 (Dale and Ow, 1990; 1991; Sauer and Henderson, 1988, each already of record), a modified Gin-gix system from bacteriophage Mu (Maeser and Kahmann, 1991, already of record). More recently and well after the priority date of the present application, others have tried with similarly limited results with respect to the following systems: the β recombinase-six system from a *Bacillus subtilis* plasmid (Diaz et al., 1999, already of record), the $\gamma\delta$ -resolvase-res system from the bacterial transposon Tn1000 (Schwikardi and Dröge, 2000, already of record, and the integrase-att system from bacteriophage λ (Lorbach et al., 2000, already of record).

None of these systems have all of the attributes of phiC31. Except for the phage λ system, the others catalyze freely reversible recombination reactions. Additionally, the β recombinase-six system catalyzes only intra, but not intermolecular recombination (Diaz et al., 1999) the $\gamma\delta$ resolvase-res system requires special DNA topology and only a mutant form of the $\gamma\delta$ resolvases was able to excise DNA from episomal molecules in mammalian cells (Schwikardi and Dröge, 2000). With the phage λ system, integrase-mediated *attB* x *attP* or *attL* x *attR*, respectively, direct the integration into or the excision out of the bacterial genome. This is similar to the phiC31 system. However, the wild type integrase was unable to function in eukaryotic cells (Lorbach et al., 2000). Only mutant integrase proteins were partially functional

in that they can perform intramolecular, but not intermolecular reactions. Using these mutant λ integrases, Lorbach et al. (2000) demonstrated only DNA inversions in recombination targets introduced into the human genome.

Moreover, assuming for the sake of argument that Crouzet did suggest the general use of prokaryotic recombinases in eukaryotes, there is no teaching anywhere in Crouzet to suggest the use in eukaryotes of the recited prokaryotic recombinases *which are not capable of mediating in the eukaryotic cell recombination between the hybrid sites formed by the recombinase*. With respect to claim 2, Crouzet does not disclose any of the recombinases set forth therein. Where Crouzet et al. do mention the use of *attB* and *attP* sequences in the production of their minicircles, it is always in the context of a plasmid with a *bacterial* origin of replication.

With respect to new claims 48, 49, 50, 52, and 54 which further set forth a transgene located between an *attR* recombination site and an *attL* recombination site, wherein said transgene is *stably* integrated into the genome of the cell. Crouzet et al. do not teach this subject matter. Crouzet et al. teach methods of *excising* their circular double-stranded DNA molecules "minicircles" by a site-specific recombination.

Moreover, assuming *arguendo* that Crouzet et al. teach *attB* and *attP* recombination sites and further inserting their genes of interest into a host genome for the sake of producing their minicircles, the genes of interest themselves would not be located between an *attR* site and an *attL* site in the host genome, but between *attB* and *attP* recombination sites so that they may be excised and recovered for subsequent use.

Independent claims 36 and 52 place the *attB* and *attP* recombination sites on different ones of the genome and a non-genomic nucleic acid. Crouzet does not disclose this arrangement of *attB* and *attP* recombination sites in their host cells.

Accordingly, as the Crouzet reference neither discloses nor enables the claimed subject matter, the Applicants respectfully request that the above rejection be reconsidered and withdrawn with respect to all the pending claims.

Response to the Rejection of claims 1 to 12, 32 to 34 and 36 to 47 over Crouzet and further in view of Thorpe et al.

In accordance with MPEP §2143.03, all the claim limitations must be taught or suggested by the alleged prior art in order to establish prima facie obviousness of a claimed invention. Thorpe does not suggest using a prokaryotic recombinase in a eukaryotic cell to obtain an irreversible site specific combination. Nor, in view of the above enablement discussion would one of ordinary skill in the art be motivated to make a combination where there would be no expectation that the combination would work.

As the combination with Thorpe does not supply the teachings absent in Crouzet, the Applicants respectfully request that the above rejection be reconsidered and withdrawn.

CONCLUSION

In view of the foregoing, Applicants believe all claims now pending in this Application merit an allowance. The issuance of a formal Notice of Allowance at an early date is respectfully requested.

If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 925-472-5000.

Respectfully submitted,



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